Molecular aspects of antibody-antigen interactions

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Summary

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Antibody molecules, produced as a response against foreign substances, interact with their antigen in a very specific manner. Antibodies with a predetermined specificity (monoclonal antibodies) can be produced and are widely used in medicine and science as indicator molecules. Genetic engineering of antibody molecules offers the possibility to study the nature of the antibody-antigen interaction in more detail and to modify antibody molecules for specific applications.

Only a small part of the antibody molecule is directly involved in antigen binding. This antigen binding site is made up of the variable domains of the heavy- and light chain. Also, only part of the antigen molecule is involved in the antibody-antigen interaction. The part of the antigen that is involved in the interaction with the antibody is referred to as the 'epitope'. For some applications size reduction of the antibody molecule could be beneficial, e.g. a more stable ligand in biosensor applications and in immunoaffinity chromatography, and for structural investigations with NMR spectroscopy.

In this thesis size reduction of both the antibody and the antigen are studied in order to identify the minimal parts of the molecules involved in the interaction. For this purpose the monoclonal antibody A16, that recognizes an epitope on glycoprotein D of herpes simplex virus, was used as a model system. Several approaches for size reduction of the A16 antibody were studied simultaneously. Antibody fragments of different size derived from the variable domains, either produced in E. coli or as synthetic peptides, were evaluated. The soluble expression of recombinant fragments proved to be very difficult. Unfortunately, neither the recombinant fragments nor the synthetic peptides showed a detectable affinity towards the antigen. Hence, antigen binding of fragments smaller than the Fv (made up of the variable domains of heavy- and light chain) is only encountered in very special cases.

In order to identify the minimal part of the antigen involved in the interaction with the antibody, A16 was used to select binding peptides from a large library of random peptides that are displayed on the surface of a filamentous bacteriophage. A binding pattern of three essential amino acid residues was identified. With the use of synthetic peptides and biosensor technology a seven residue peptide was identified as the minimal antibody binding site. A peptide derived from the library displayed similar binding- and immunological characteristics as a peptide derived from the antigen (glycoprotein D). This method failed to select binding peptides

using the non-antibody target myoglobin. During these studies a method was developed to study the binding characteristics of selected phage clones, prior to DNA sequencing and peptide synthesis, using biosensor technology.